Sentinel node in oral cancer: the nuclear medicine aspects: a survey from the sentinel european node trial

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Abstract: PURPOSE: Nuclear imaging plays a crucial role in lymphatic mapping of oral cancer. This evaluation represents a subanalysis of the original multicenter SENT trial data set, involving 434 patients with T1-T2, N0, and M0 oral squamous cell carcinoma. The impact of acquisition techniques, tracer injection timing relative to surgery, and causes of false-negative rate were assessed. METHODS: Three to 24 hours before surgery, all patients received a dose of Tc-nanocolloid (10-175 MBq), followed by lymphoscintigraphy. According to institutional protocols, all patients underwent preoperative dynamic/static scan and/or SPECT/CT. RESULTS: Lymphoscintigraphy identified 723 lymphatic basins. 1398 sentinel lymph nodes (SNs) were biopsied (3.2 SN per patient; range, 1-10). Dynamic scan allowed the differentiation of sentinel nodes from second tier lymph nodes. SPECT/CT allowed more accurate anatomical localization and estimated SN depth more efficiently. After pathological examination, 9.9% of the SN excised (138 of 1398 SNs) showed metastases. The first neck level (NL) containing SN+ was NL I in 28.6%, NL IIa in 44.8%, NL IIb in 2.8%, NL III in 17.1%, and NL IV in 6.7% of positive patients. Approximately 96% of positive SNs were localized in the first and second lymphatic basin visualized using lymphoscintigraphy. After neck dissection, the SN+ was the only lymph node containing metastasis in approximately 80% of patients. CONCLUSIONS: Best results were observed using a dynamic scan in combination with SPECT/CT. A shorter interval between tracer injection, imaging, and surgery resulted in a lower false-negative rate. At least 2 NLs have to be harvested, as this may increase the detection of lymphatic metastases.

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Sentinel Node in Oral Cancer

The Nuclear Medicine Aspects. A Survey from the Sentinel European Node Trial

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Purpose: Nuclear imaging plays a crucial role in lymphatic mapping of oral cancer. This evaluation represents a subanalysis of the original multicenter SENT trial data set, involving 434 patients with T1-T2, N0, and M0 oral squamous cell carcinoma. The impact of acquisition techniques, tracer injection timing relative to surgery, and causes of false-negative rate were assessed.

Methods: Three to 24 hours before surgery, all patients received a dose of ^{99m}Tc-nanocolloid (10–175 MBq), followed by lymphoscintigraphy. According to institutional protocols, all patients underwent preoperative dynamic/static scan and/or SPECT/CT.

Results: Lymphoscintigraphy identified 723 lymphatic basins. 1398 sentinel lymph nodes (SNs) were biopsied (3.2 SN per patient; range, 1–10). Dynamic scan allowed the differentiation of sentinel nodes from second tier lymph nodes. SPECT/CT allowed more accurate anatomical localization and estimated SN depth more efficiently. After pathological examination, 9.9% of the SN excised (138 of 1398 SNs) showed metastases. The first neck level (NL) containing SN+ was NL I in 28.6%, NL IIa in 44.8%, NL IIb in 2.8%, NL III in 17.1%, and NL IV in 6.7% of positive patients. Approximately 96% of positive SNs were localized in the first and second lymphatic basin visualized using lymphoscintigraphy. After neck dissection, the SN+ was the only lymph node containing metastasis in approximately 80% of patients.

Conclusions: Best results were observed using a dynamic scan in combination with SPECT/CT. A shorter interval between tracer injection, imaging, and surgery resulted in a lower false-negative rate. At least 2 NLs have to be harvested, as this may increase the detection of lymphatic metastases.

Key Words: squamous cell carcinoma, sentinel lymph nodes, lymphoscintigraphy, single-photon emission computed tomography, head and neck cancer, gamma probe, sentinel lymph node biopsy, lymphatic metastasis, neck dissection

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TABLE 1. Nuclear Medicine Procedures Applied

Procedures	Tracer	% of Patients
Radiopharmaceutical	Nanocolloid	91%
	NanoCis	9%
Same day Dose 47 MBq (10–94)		64%
Two day Dose 70 MBq (20–175)		36%
Volume mL 0.4 (0.2–1)		100%
Tracer injections 4 (2–6)		100%
Peritumoral injection		100%
Skin mark		93%
Timing	Same day	64%
	Two days	36%
Gamma Probe		100%
Mobile Camera		10%
3D Navigation		10%
Blue-dye		29%
Gamma camera		100%
Zoom	x1.5	11%
	x1.0	89%
Collimator	LEAP	11%
	LEHR	89%
Dynamic		78%
Frames	30”–60” in total 15’– 30’	100%
Matrix	128 × 128	100%
STATIC		45%
Preset time	120”–300”	82%
	600”	18%
Matrix	256 × 256	100%
SPECT		46%
Degree of rotation	360°(180°x2)	100%
Number of views	60*	35%
	32 **	33%
	120 ***	33%
Time per view	15” *	35%
	30”–40” **	33%
	8” ***	33%
Orbit	Circular * ***	67%
	Noncircular **	33%
SPECT/CT		29%

The presence of lymph node metastases is generally the most important prognostic factor in head and neck cancers.[1] In oral squamous cell carcinoma (OSCC), the prevalence of occult metastases ranges from 15% to 35%.[2] Unfortunately, even highly sophisticated radiological imaging (CT, MRI, US, and PET/CT) is unable to reliably detect or exclude small cervical metastases.[3]

Historically, management of T1-T2N0 OSCC was to perform an elective lymph node neck dissection (END),[4–7] but this approach may result in overtreatment in up to 75% of patients. The wait-and-see policy is not recommended because of the risk of advanced late metastases and difficult salvage surgery.[8,9] In a recent landmark paper comparing END to wait and see in a prospective randomized trial, the patients undergoing END showed a clear survival benefit.[10] There is a need to improve the detection of occult lymph node metastases to offer END in only those patients that will benefit from the procedure.[11]

Sentinel node biopsy (SNB) attempts to identify the regional lymph nodes most likely to harbor metastasis. The sentinel node (SN) is any lymph node or group of lymph nodes, which receives lymphatic drainage directly from the primary tumor. Sentinel nodes may not necessarily be the ones closest to the tumor, and usually, there is more than one SN. All other lymph nodes are reached after passing through the sentinel node and are therefore called second-tier or second echelon lymph nodes.[12]

Sentinel node biopsy is universally recognized as an accurate staging method in melanoma and breast cancer, reliably selecting patients for lymph node dissection. In the 1990s, Pitman first used intraoperative lymphatic mapping with isosulfan blue dye for the detection of SNs in OSCC.[13] The first successful SNB in OSCC using a gamma probe was reported in 1996 by Alex and Krag.[14] Since then, an increasing number of validation studies and observational trials have proven the applicability of lymphoscintigraphy and SNB in OSCC with high sensitivity, negative predictive value, and low regional failure rate in patients with a negative SNB.[15–25]

From 2005 to 2010, a prospective European Sentinel Node observational Trial (SENT) involving 14 European institutions was conducted to establish the role of SNB in the management of early OSCC.[26] Sentinel nodes were mapped using lymphoscintigraphy, requiring a multidisciplinary approach involving nuclear medicine physicians, surgeons, and pathologists in a variety of practice settings.

The aim of this paper is to analyze the role and compare methods of lymphoscintigraphy for lymphatic mapping and identification of the SN in OSCC in the SENT multicenter trial.

PATIENTS AND METHODS

From October 2005 to October 2010, approximately 482 consecutive patients with cT1-T2cN0 OSCC were recruited from 14 European centers in the SENT study. The study was approved by the EORTC (No. 24021) protocol review committee in accordance with local research ethics committee approval. This evaluation represents a subanalysis of the original SENT data set with emphasis on lymphoscintigraphic data. Forty-eight patients had inadequate data and were excluded from the analysis, leaving 434 eligible cases; some patients that did not follow the SENT protocol and thus excluded from long-term follow-up analysis were included in this nuclear medicine evaluation.

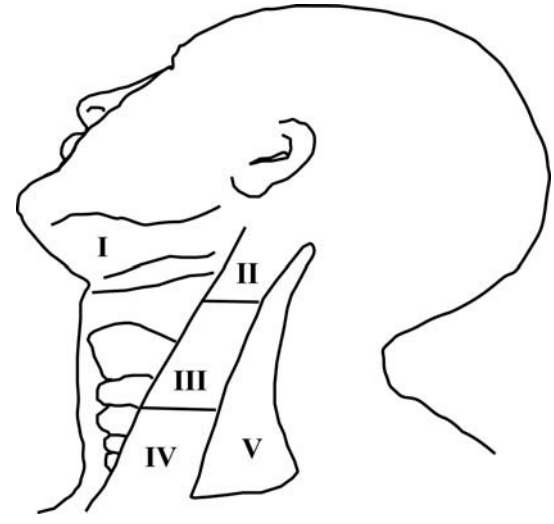

FIGURE 1. Neck levels according to American Academy of Otolaryngology–Head and Neck Surgery (picture homemade).

TABLE 2. Tumor Location and SENTINEL Node Status

Site of Primary	No. Patients (%)	SNB– (%)	SNB+ (%)
Anterior 2/3 tongue	218 (50.2)	158 (72.5)	60 (27.5)
Posterior 1/3 tongue	44 (10.1)	28 (63.6)	12 (27.4)
Buccal mucosa	19 (4.4)	15 (78.9)	4 (21.1)
Soft palate	10 (2.3)	8 (80.0)	2 (20.0)
FOM	110 (25.4)	91 (82.7)	19 (17.3)
Retro-molar	9 (2.1)	6 (66.7)	3 (33.3)
Lower alveolus	8 (1.8)	4 (50.0)	4 (50.0)
Upper alveolus	7 (1.6)	6 (85.7)	1 (14.3)
Lower lip	6 (1.4)	6 (100.0)	0 (0.0)
Hard palate	3 (0.7)	3 (100.0)	0 (0.0)
Total	**434 (100.0)**	**329 (75.8)**	**105 (24.2)**

All patients were staged N0 based on CT or MRI. Any lymph nodes larger than 1.5 cm in level II and larger than 1 cm in all other levels or lymph nodes with a round shape, central necrosis, and peripheral contrast enhancement were considered as pathological and therefore excluded the patient from participation. No previous neck pathology or treatment to the primary tumor or neck that could alter lymphatic drainage channels had been performed.

All patients underwent injection of radiotracer and lymphoscintigraphy imaging between 3 and 24 hours before surgery according to local institutional protocols. Any jewelry, dental prosthesis, and all relevant metallic items were removed. The position mirrored that of the patient at surgery. The radiopharmaceuticals used in our study were ^{99m}Tc-HSA Nanocoll (GE Healthcare) in 395 (91%) of 434 patients and ^{99m}Tc-Nanocis (CIS Bio) in 39 (9%) of 434 patients. If necessary, a local anesthetic (10% lidocaine spray) was given before injection to reduce patient discomfort. The average dose of injected radioactivity was 47 MBq (range, 10–94 MBq) for a same-day protocol and 70 MBq (range, 23–175 MBq), using a 2-day protocol, respectively.

The radiotracer was diluted in a small volume of saline solution, ranging from 0.2 to 1 mL.

The radiotracer was injected submucosally in 4 sites around the tumor in an attempt to completely surround the border of the tumor with the radiocolloid. A mouthwash was used immediately after injection to prevent pooling or swallowing of tracer.

Lymphoscintigraphy acquisition started within a few minutes of tracer injection and was initially monitored dynamically with the gamma camera in the anterior view in 338 (78% extra) of 434 patients.

Dynamic images for 5'–30' (15"–30" frames; matrix, 128 × 128) were acquired with a gamma camera equipped with a LEAP or LEHR collimator, minimizing the distance from the collimator face to the patient, to increase spatial resolution. The energy window of 20% was centred at 140 KeV, zoom ×1.5, 128 × 128 or 256 × 256 matrix.

In 195 (45%) of 434 patients, when accumulation of the radiotracer in the first node(s) occurred, an early planar static scan (256 × 256, preset time 300"–600", zoom ×1.0–1.5, collimator LEAP or LEHR) was taken, followed by a lateral and/or oblique view depending on the lymphatic drainage seen. The SNs were localized with the gamma camera using an external radioactive marker (^{99m}Tc or ^{57}Co) or with a flood-field source in anterior and lateral projections. The skin overlying the SNs was marked with an indelible marker pen in 404 (93%) of 434 patients to help the surgeons localize the SN during surgery. SPECT imaging was performed in 200 (46%) of 434 patients. SPECT acquisition parameters are described in Table 1.

A SPECT/CT system was used in 126 (29%) of 434 cases.

Unexpected lymphatic drainage patterns including unilateral level IV or V occurred in some tumors.

The surgeon used a hand-held gamma probe to detect the SN in the area of the skin marks to direct the skin incision and to perform a radioguided SNB intraoperatively. Radioactivity was confirmed in the excised SN, and the cutoff value for an SN was at least 3 times the background activity. After excision of all SNs, the level of radioactivity in situ had to drop to background levels to ensure excision of all SNs.

In 127 (29%) of 434 patients, additional intraoperative peritumoral injections of Patent Blue V dye (~0.5 mL) were performed in an identical manner to the injections of radiotracer. SNs were labeled according to their radioactivity count and anatomical neck level according to the a classification system developed by the American Academy of Otolaryngology–Head and Neck Surgery (Fig. 1).[27]

Sentinel nodes were histologically evaluated with step-serial sectioning, staining with hematoxylin and eosin and immunohistochemistry with a pan cytokeratin antibody AE1/3 according to the modified Canniesburn protocol.[28] Demographic data, site of primary tumor, lymphatic drainage patterns, anatomical location, and number and order of appearance at lymphoscintigraphy of SNs were collected for each patient. The collected data were subjected to univariate statistical analysis.

RESULTS

In the SENT trial, 434 patients with OSCC staged cT1/T2cN0 (257 male and 177 female patients) with a mean age of 60.8 years (range, 28–92 years) were eligible for the subanalysis of nuclear medicine data.

The sites of the primary tumors and the results of the SNB are shown in Table 2.

Tumor localization was well lateralized in 386 (89%) of 434 patients. The primary tumor was found on the right side in 199 (46%) of 434 patients, left side in 187 (43%) of 434 patients, and in the midline in 48 (11%) of 434 patients, respectively. On lymphoscintigraphy, the lymphatic drainage was unilateral in 361 (83%) of 434 patients and bilateral in 73 (17%) of 434 patients.

Unexpected lymphatic drainage patterns were observed in 69 (16%) of 434 patients.

TABLE 3. Lymphoscintigraphic Order of Appearance of SN According to Anatomical Neck Level (723 Lymphatic Basins Dissected in 434 Patients)

Lymphatic Basins	NL I	NL IIa	NL IIb	NL III	NL IV	NL V	N/A	Total
Main basin	124 (28.6)	234 (53.9)	10 (2.3)	58 (13.4)	6 (1.4)	2 (0.4)	0 (0.0)	**434 (59.9)**
Second basin	36 (15.1)	61 (25.5)	12 (5.0)	94 (39.3)	19 (7.9)	11 (4.6)	6 (2.5)	**239 (33.1)**
Third basin	6 (13.0)	12 (26.1)	1 (0.2)	10 (21.7)	17 (36.9)	0 (0.0)	0 (0.0)	**46 (6.4)**
Fourth basin	0 (0.0)	1 (25.0)	0 (0.0)	1 (25.0)	0 (0.0)	2 (50.0)	0 (0.0)	**4 (0.6)**
Total	**166 (22.9)**	**308 (42.6)**	**23 (3.2)**	**163 (22.5)**	**42 (5.8)**	**15 (2.0)**	**6 (1.0)**	**723 (100)**

TABLE 4. Location of SNs According to Appearance on Dynamic Lymphoscintigraphy

#SN/Neck Level	#SN Main Basin	#SN Second Basin	#SN Third Basin	#SN Fourth Basin	#SN Total
NL I	291 (15.7)	70 (5.0)	9 (0.6)	0 (0.0)	**370 (26.5)**
NL IIa	481 (34.4)	106 (7.6)	14 (1.0)	2 (0.1)	**603 (43.1)**
NL IIb	18 (1.3)	19 (1.4)	1 (0.1)	0 (0.0)	**38 (2.7)**
NL III	104 (7.4)	157 (11.2)	13 (0.9)	2 (0.1)	**276 (19.7)**
NL IV	17 (1.2)	29 (2.1)	22 (1.6)	0 (0.0)	**68 (4.8)**
NL V	7 (0.5)	20 (1.4)	0 (0.0)	4 (0.4)	**31 (2.2)**
N/A	0 (0.0)	12 (0.8)	0 (0.0)	0 (0.0)	**12 (1.0)**
Total	**918 (65.7)**	**413 (29.5)**	**59 (4.2)**	**8 (0.6)**	**1398 (100.0)**

In particular, bilateral lymphatic drainage was observed in 47of 386 patients with a well-lateralized tumor, and conversely, unilateral lymphatic drainage was observed in 22 of 48 patients with a midline tumor.

Using dynamic scintigraphy, a primary lymphatic basin was identified in all patients in a mean time of 5 minutes (range up to 120). In 145 (33%) of 434 patients, there was just one basin identified. Two lymphatic basins were identified in 239 (55%) of 434 patients, in a mean time of 15 minutes (range up to 120). Three lymphatic basins were identified in 46 (11%) of 434 patients, in a mean time of 16 minutes (range up to 120). Four lymphatic basins were identified in 4 (1%) of 434 cases.

Table 3 describes the anatomical location (NL) of the lymphatic basins as assessed using lymphoscintigraphy.

In total, 723 lymphatic basins were dissected with a mean of 1.7 (range, 1–4) lymphatic basins per patient. The classification of first, second, third, or fourth lymphatic basins was based on the timing of tracer appearance and intensity of uptake.

The average number of SNs removed per lymphatic basin was 1.9 (range, 1–5). A total of 1398 radioactive SNs were removed in 434 patients, resulting in a mean of 3.2 SNs per patient (range, 1–10). In the 73 patients with bilateral lymphatic drainage, the mean number of SNs was 3.9 (range, 2–10). The distribution of the SNs within the neck levels (NL) is described in Table 4.

Only in 1 (0.2%) of 434 patients was the lymphoscintigraphy technique ineffective and did not demonstrate an SN. After pathological examination, 138 (9.9%) of 1398 SNs showed metastases. The 135 positive SNs upstaged 105 (24.2%) of 434 patients. The anatomical localization of positive SNs is shown in Table 5. Approximately 108 (78%) of 138 positive SNs were localized in the main lymphatic basin, allowing identification of 89 (85%) of 105 positive patients. Approximately 24 (17%) of 138 positive SNs were localized in the second lymphatic basin, allowing identification of 12 (11%) of 105 positive patients. The remaining 6 (4%) of 138 positive SNs were found in the third lymphatic basin, upstaging 4 (4%) of 105 patients. No positive tumor deposits were found in the fourth lymphatic basin.

Neck dissection (ND) was performed in all 105 patients with positive SNB. After ND, the positive SNs remained the only lymph node containing metastases in 84 (80%) of 105 patients. In 127 (29%) of 434 patients, lymphoscintigraphy was combined with intraoperative injection of blue dye, and in this group, 392 radioactive SNs were harvested. The SNs were "hot and blue" in 186 (47.4%) of 392 cases, and only "hot" in 206 (52.6%) of 392 cases. The mean number of SNs per patient removed with and without the use of blue dye was 3.1 versus 3.2, respectively. This difference was not statistically significant ($P = 0.191$).

The mean number of lymphatic basins biopsied was 1.6 in the 1-day protocol 276 (64%) of 434 patients and 1.8 in the 2-day protocol 158 (36%) of 434 patients. Similarly, the mean number of SNs harvested per patient was 3.20 and 3.24. All these differences were not statistically significant ($P = 0.240$).

When considering if the number of lower echelon neck levels were increased by a 2-day protocol, we found that biopsies were taken from level IV and V in 6.3% of 1-day patients compared with 10.3% in 2-day patients ($P = 0.498$). Using a same-day protocol, the false negative rate (FNR, percentage of cases in which a test fails to detect the disease) was 10.8%, compared with 13.3% in the 2-day protocol ($P = 0.265$) (Table 6).

SPECT imaging identified a mean of 3.29 SNs per patient compared with 3.14 by planar lymphoscintigraphy. The FNR was 10.0% compared with 13.5% without SPECT ($P = 0.297$) (Table 7).

DISCUSSION

Sentinel node biopsy in OSCC is rapidly evolving after innovation of new technologies. In the neck, there are normally more than 300 lymph nodes, and sentinel nodes are often in proximity to the injection area. Embedded in the epithelium of the oral cavity mucosa, there are mainly blind ending lymphatic capillaries connected to an underlying lymphatic plexus, which channels lymph into deeper lymphatic vessels. These are then directed into the regional lymph nodes.

TABLE 5. Anatomical Localization, Number and Order of Tracer Appearance of Positive SNs in105 Patients With Lymphatic Metastases

#SN+/Neck Level	#SN+ Main Basin	#SN+ Second Basin	#SN+ Third Basin	#SN+ Total	First Neck Level Containing SN+
NL I	32 (23.2)	4 (2.9)	0 (0.0)	**36 (26.1)**	**30 (28.6)**
NL IIa	55 (39.9)	8 (5.8)	0 (0.0)	**63 (45.7)**	**47 (44.8)**
NL IIb	2 (1.4)	1 (0.7)	0 (0.0)	**3 (2.1)**	**3 (2.8)**
NL III	14 (10.1)	9 (6.5)	2 (1.4)	**25 (18.1)**	**18 (17.1)**
NL IV	5 (3.6)	2 (1.4)	4 (2.9)	**11 (8.0)**	**7 (6.7)**
Total	**108 (78.3)**	**24 (17.4)**	**6 (4.3)**	**138 (100.0)**	**105 (100.0)**

TABLE 6. Comparison of Results Between Same Day and 2-Day Protocol

Protocol	#Patients	#Lymphatic Basin/Patient	Neck Level Involved %				SN/Patient	#SN−	#SN+	True Positive	True Negative	False-Negative	False-Negative Rate %
			I	IIa IIb III	IV V	N/A							
Same day	276 (63.6)	1.6	23.3	69.6	6.3	0.7	3.20	806 (91.1)	79 (8.9)	66 (15.2)	202 (46.5)	8 (1.8)	10.8
Two days	158 (36.4)	1.8	22.3	66.3	10.3	1.1	3.24	454 (88.5)	59 (11.5)	39 (8.9)	113 (26.0)	6 (1.4)	13.3
Total	**434 (100.0)**	**1.7**	**22.9**	**68.3**	**7.8**	**1.0**	**3.2**	**1260 (91.1)**	**138 (9.9)**	**105 (24.2)**	**315 (72.6)**	**14 (3.2)**	**11.7**
								1398 (100.0)			434 (100.0)		

TABLE 7. Comparison of Sentinel Node Imaging Procedures

NM Procedure	#Patients	#Lymphatic Basin/Patient	Neck Level Involved %				SN/Patient	#SN−	#SN+	True Positive	True Negative	False-Negative	False-Negative Rate %
			I	IIa IIb IIII	IV V	N/A							
Dynamic/static	241 (55.5)	**1.7**	16.5	71.9	8.9	0.0	3.24	716 (91.8)	64 (8.2)	51 (11.8)	182 (39.2)	8 (1.8)	13.5
Dynamic/SPECT	193 (44.5)	**1.6**	25.7	65.6	7.7	1.2	3.29	544 (88.0)	74 (11.9)	54 (12.4)	133 (33.4)	6 (1.4)	10.0
Total	**434 (100.0)**	**1.7**	**22.9**	**68.3**	**7.8**	**1.0**	**3.2**	**1260 (91.1)**	**138 (9.9)**	**105 (24.2)**	**315 (72.6)**	**14 (3.2)**	**11.7**
								1398 (100.0)			**434 (100.0)**		

Sometimes, there is more than one lymphatic channel originating in the region of the primary cancer and running to different lymphatic basins. Because of this complex arrangement, an experienced team is required for optimal identification of SN in the head and neck.

Solitary use of the gamma probe for SN identification is discouraged because it samples only one particular area for a short time, and its sensitivity is lower than the gamma camera. Accurate lymphatic mapping is crucial for correct identification of SNs, considering the individual and unpredictable lymphatic drainage patterns of OSCC.[29–31]

Minimal requirements for adequate SNB are the use of peritumoral radiotracer injection, preoperative lymphoscintigraphic scanning and the use of a peroperative gamma probe.

These instruments allow for reliable lymphatic mapping and SN localization and identification.

Selection of the radiotracer, availability of dynamic/static and SPECT-CT lymphoscintigraphy, and type of gamma probe are dependent on individual institutions' resources. Many factors influence correct identification of SNs in OSCC.

An average activity of 47 MBq was injected for a same-day protocol, whereas an average activity of 70 MBq was injected with a 2-day protocol. The injection technique of tracer can impact on the success rate of SNB. Intratumoral or deeper injection should be avoided because intense bleeding at injection points may increase background activity with low-quality images resulting in difficult identification of SNs. Superficial perilesional injections of radiotracer are preferred to intratumoral or deeper injections because the lymphatic capillary plexuses in the mucosa provide a larger surface area for uptake with a faster lymph drainage and a better identification of lymph collecting vessels and SN in a shorter time.[32–34]

Colloidal radiopharmaceuticals are phagocytosed by the macrophages within the lymph node, retaining the tracer in the draining node. The higher signal-to-noise ratio facilitates gamma probe identification of the SN. Normally, if the injection technique is correct, the main lymphatic basin is demonstrated by dynamic imaging within a few minutes.

The value of blue dye injection is debatable. Data from the SENT trial confirm a low concordance between blue dye and lymphoscintigraphy. Blue dye and nanocolloids have different pharmacokinetic and pharmacodynamic properties, and therefore, results differ as well. Blue dye moves rapidly downstream to distal lymph nodes, whereas nanocolloids are more efficiently trapped, achieving a superior identification rate.

Planar lymphoscintigraphy is routinely used for preoperative sentinel node localization. Gamma cameras have the advantage of simultaneously sampling multiple lymph node basins in the entire neck, with improved staging. A LEAP/LEGP collimator, which increases the sensitivity of the gamma camera, and an adequate zoom factor ($\times 1.5$) may be able to demonstrate lymphatic drainage pathways and SNs closely related to the primary tumor (Fig. 2).

Dynamic planar imaging in the anterior view may show individual lymph drainage pathways demonstrating the direction of lymphatic drainage. In our trial, 96% of positive SNs were localized at the main or second tier lymphatic basin, confirming the importance of this approach.

Static planar images in lateral/oblique views show an overview of the number and localization of SNs. The levels of these SNs can be further localized using external radioactive markers, such as a ^{99m}Tc or a ^{57}Co-source pen for marking the overlying skin or a ^{57}Co flood source transmission scan.

Lymphoscintigrams demonstrated the large variability and complexity of lymphatic drainage patterns in OSCC. The main lymphatic basin is specific for each patient regardless of the site of primary tumor. This personalized lymphatic mapping reveals an unexpected lymphatic drainage pattern in 16% of patients with contralateral drainage in well-lateralized tumors or unilateral drainage in midline tumors. This considerable number confirms the importance of lymphoscintigraphy before surgery. This unexpected drainage would not have been sampled by routine END.

In some patients, level I, IIa, or III lymph nodes can be connected with the primary cancer in series or directly by an individual lymphatic collector (Fig. 3). This may explain why, in some cases, metastases can bypass nodes near the primary cancer and can be found in a higher (often more caudal) neck level. However, the shine through phenomenon may simulate this finding.

SPECT was performed in addition to planar images with the principal purpose of anatomical localization of SNs already identified using lymphoscintigraphy.

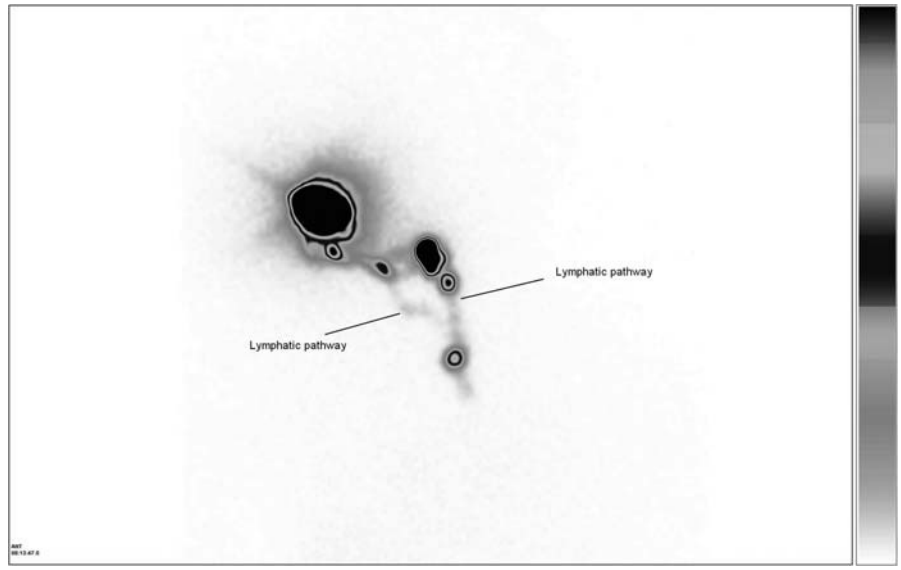

FIGURE 2. SCC of tongue 2/3 anterior: the early scan in the left lateral view shows the injection point and the lymphatic drainage way directed toward a SN at NL IL; from this SN, we can see 2 lymphatic ways directed toward the NL IIa and the NL III. The distal lymph node (NL III) seems to be connected with cancer area in series and in parallel.

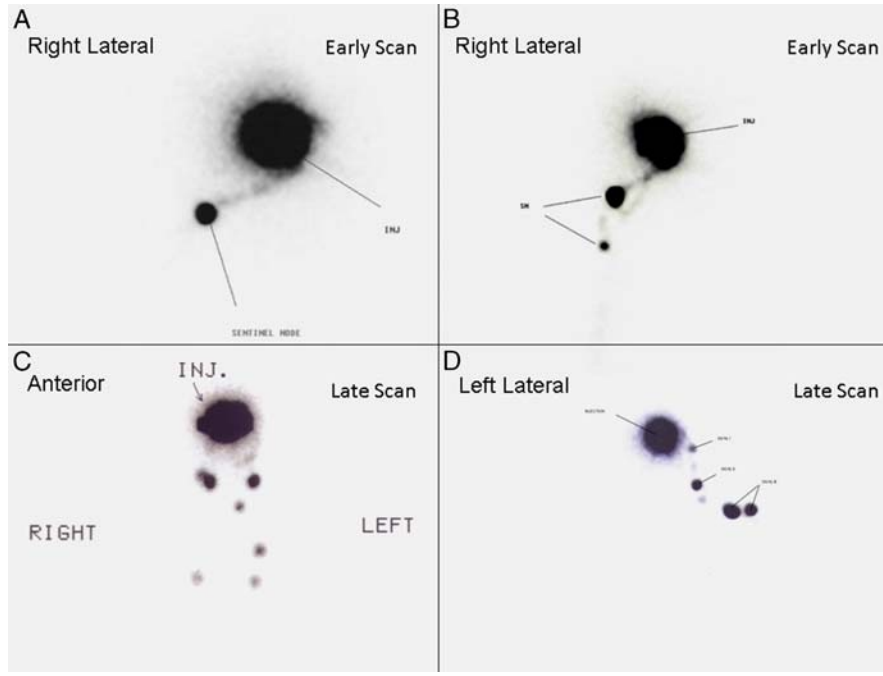

FIGURE 3. Images show different lymphatic drainage patterns, in 4 patients with an SCC of the oral cavity. **A,** early scan (5′) shows a single lymphatic way directed toward a SN localized at NL IIa (right side). **B,** Early scan (5′) shows 2 lymphatic ways directed toward a SN localized at NL IIa and toward a SN localized at NL III (right side). **C,** Late scan (30′) in anterior view shows a bilateral lymphatic drainage with SNs and second tier lymph nodes at lower neck levels. **D,** Late scan (30′) in the left lateral view shows the SNs at NLs I, II, and III.

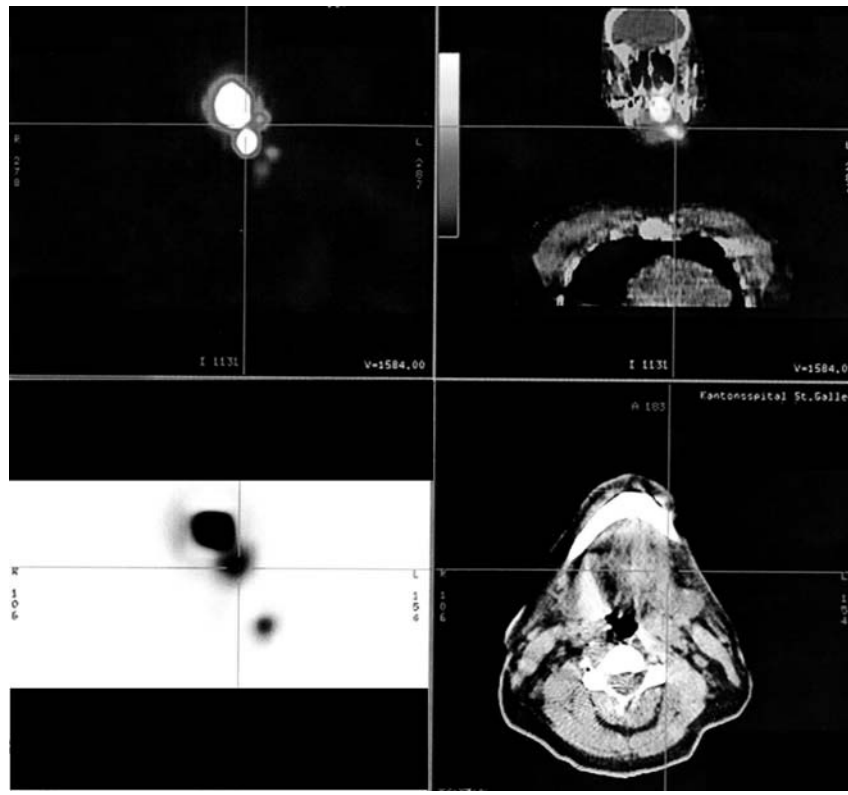

FIGURE 4. SPECT-CT imaging in OSCC: the image shows a SN at NL I.

For more accurate lymphatic mapping, an early scan with oblique views and/or SPECT is required to identify SNs that are closely related to the primary cancer. An interesting trend was found using SPECT imaging: 25.7% of level 1 nodes were biopsied, instead of 16.5% found in the group of patients studied without SPECT. In addition, in the group of cases studied with SPECT, the FNR was 10% compared with 13.5% in the group of patients studied without SPECT. These findings may also be attributed to the shine through phenomenon, through which NL I SNs can be difficult to identify.

In floor-of-mouth cancer, it is suggested that the primary tumor is excised before performing SNB to reduce the shine-through phenomenon as radioactivity at the primary site may obscure SN located at NL I.[35]

Fused SPECT/CT imaging may allow more accurate anatomical localization of the SNs adjacent to the primary lesion (Fig. 4). Sometimes, SPECT/CT may result in more visible foci and, therefore, more SNs. If these are in greater proximity to the primary tumor site, the additional SNs may have clinical relevance.[36–39]

Dynamic scan is recommended to identify the velocity from the injection site to the first echelon node as well as the appearance of the other echelon-nodes. The association with SPECT/CT acquisition following the static planar imaging appears to be very useful to better lobalize the site of the sentinel node(s) as suggested by current EANM-SENT joint practice guidelines and by a recent Phase III Multi-institutional Trial on OSCC.[33,40]

Variable intervals of time between tracer injection, imaging, and surgery were used in this study. With regard to the timing of surgery, we observed that 63.6% of patients had a same-day protocol, whereas 36.4% had a 2-day protocol.

The same-day protocol resulted in a high percentage of lymphatic basins being dissected closely to the primary (I-III NLs) of 93.2% as compared with 88.6% using a 2-day protocol. There was no statistically significant difference between the 2 groups regarding the mean number of lymphatic basins biopsied and the mean number of SNs harvested per patient. Using a same-day protocol, the FNR was 10.8%. Using a 2-day protocol, we observed 10.3% of lower NLs (IV, V) biopsied versus 6.3% with a same-day protocol. This approach resulted in an FNR of 13.3%. The radiotracer may appear progressively with time in second-tier lymph nodes that have a lower incidence of metastases. The same-day protocol may be a more advantageous approach because fewer nonrelevant SNs were sampled, although no significant differences were found.

In patients with neck dissections, SNs were the only lymph nodes containing metastasis in approximately 80% of positive patients, confirming the high sensitivity of SN sampling and the assumption that a primary tumor drains to relatively few lymph nodes before disseminating to the remaining local nodal field.

In our trial, we observed that 96% of positive SNs were localized in the first and second lymphatic basin as visualized using dynamic scanning. The anatomical localization of 92% of positive SNs were in I, IIa, IIb, and III NLs. For these reasons, accurate lymphatic mapping and perioperative probe scanning of these NLs is always recommended.

New technologies such as a mobile camera or 3D navigation were used in only 10% of cases.

These new technological approaches may aid intraoperative identification of SNs. Freehand SPECT is a 3D tomographic nuclear imaging modality based on the concepts of SPECT, which can be used for intraoperative visualization of SNs to facilitate their localization and removal during surgery.[41]

Indocyanine green (ICG) fluorescence for SN biopsy has been used in relatively few cases, and its role is under study. The use of ICG in combination with ^{99m}Tc (hybrid tracer) may provide additional information to detect SNs in head and neck cancer.[42,43]

In conclusion, SNB is the most advanced, multidisciplinary and continuously evolving strategy available to reduce the risk from occult lymph node metastases in cT1/T2N0 OSCC patients and may play a crucial role in the decision on whether to perform neck dissection in clinical practice.

From this study, it can be concluded that a dynamic planar imaging is essential to differentiate between SNs and second echelon nodes; SPECT/CT may be helpful in anatomical localization of the SNs. A same-day protocol may be better able to identify only the most relevant (sentinel) lymph nodes. The value of blue dye in addition to radiolabeled nanocolloids is limited. At the least, biopsy of SNs in the first and second lymphatic basin appears to be necessary.

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